CLAIMS

What is claimed is:

1. A de novo synthesized plasmid comprising at least a replication origin and a selection marker gene wherein;

(a) the replication origin contains sequences relevant to autonomous plasmid replication in a host cell; and

(b) the selection marker gene contains sequences relevant to the selection of a plasmid in a host cell.

2. The plasmid according to claim 1, wherein the plasmid is not modified from the plasmid previously obtained from natural sources.

3. The plasmid according to claim 1, wherein the plasmid is not modified from the plasmid previously obtained from recombinant sources.

4. The plasmid according to claim 1, wherein the replication origin allows the autonomous plasmid replication in a host cell.

5. The plasmid according to claim 1, wherein the selection marker gene encodes a product indicative of plasmid maintenance in a host cell.

6. A method of preparing a de novo synthesized plasmid combined from at least two DNA fragments comprising:

(a) preparing a linear replication origin DNA fragment;

(b) preparing a linear selection marker gene DNA fragment;

(c) combining the DNA fragments prepared from steps (a) and (b) to form a circular de novo synthesized plasmid;

(d) introducing the plasmid made from step (c) into a host cell; and

(e) selecting the plasmid with appropriate replication origin and selection marker from transformed host cells.

7. The method according to claim 6, wherein any DNA fragment alone used for combining the de novo synthesized plasmid cannot confer both autonomous DNA replication and selection to a plasmid.

8. The method according to claim 6, wherein the linear DNA fragments of steps (a) and (b) are prepared from polymerase chain reaction.

9. The method according to claim 6, wherein the linear DNA fragments of steps (a) and (b) are prepared from restriction digestion.

10. A method of using a de novo synthesized plasmid comprising:

(a) Linearizing the de novo synthesized plasmid;

(b) inserting one or more functional DNA fragments to the linearized plasmid to make other plasmids;

(c) introducing the plasmids made from step (b) into host cells;

(d) selecting the plasmids and host cells with desired properties; and

(e) using the plasmids and host cells for biomedical applications.

11. The method according to claim 10, wherein linearizing the plasmid was achieved by restriction digestion.

12. The method according to claim 10, wherein linearizing the plasmid was achieved by PCR.

13. The method according to claim 10, wherein the functional DNA fragments encode a promoter, a regulatory sequence, a ribosome binding site, restriction sites, a terminator, a polypeptide, a replication origin, and a selection marker gene.

14. The method according to claim 10, wherein the desired properties are plasmid replication, selection, and the properties added by functional DNA fragments inserted from step (b).

15. The method according to claim 10, wherein the biomedical applications are DNA cloning, DNA amplification, gene expression, gene therapy, and DNA immunization.